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Primary Carcinoma of the Fallopian Tube: Comparative Genomic Hybridization Reveals High Genetic Instability and a Specific, Recurring Pattern of Chromosomal Aberrations

Kerstin Heselmeyer, Ph.D., Ann-Cathrin Hellström, M.D., Harald Blegen, M.D., Evelin Schröck, M.D., Claes Silfverswärd, M.D., Keerti Shah, M.D., Gert Auer, M.D., and Thomas Ried, M.D.

Summary: Primary fallopian tube carcinoma (PFTC) is a rare and highly aggressive tumor. Twelve cases of PFTC (stages IA to IV) were analyzed by comparative genomic hybridization. The most consistent DNA gain was mapped to chromosome arm 3q in 11 of 12 cases. In six cases, the gain of 3q was present as a high level copy number increase (amplification) with a consensus region mapped to 3q26.2-qter. In the 12 cases, other frequent gains were located on chromosome arms 1q (in 11 cases), 2q (in 10), 7q (in 9), 8q (in 9), 5p (in 8), 6p (in 7), 12p (in 7), and 14q (in 6). Frequent copy number losses occurred on chromosome arms 16q (in 8 cases), 22q (in 7), 6q (in 6), 8p (in 6), 18q (in 6), Xq (in 6), 1p (in 5), and 17p (in 5). All chromosomes were involved in chromosomal aberrations and the average number of copy alterations per case was 19.7. None of the 12 carcinomas revealed the presence of human papillomavirus (HPV) genomes. All of the cases exhibited crude aneuploidy. Strong p53 immunoreactivity could be observed in 10 of 12 cases while p21/WAF1 expression was low or undetectable. These results indicate that PFTC is a genomically highly unstable cancer, an observation that is in agreement with the poor prognosis associated with this tumor. A high frequency of 3q-gains has also been observed in HPV-related carcinomas of the uterine cervix. However, none of the PFTC was HPV-related, suggesting that the 3q-gain is independent from HPV DNA. **Key Words:** Fallopian tube—Cancer—Chromosome aberrations—Comparative genomic hybridization (CGH)—p53—DNA ploidy—Proliferation—Human papillomavirus (HPV).

Primary carcinoma of the fallopian tube (PFTC) is a rare tumor, accounting for ~ 0.15 to 1.8% of all gynecologic cancers (1–4). An incidence of 2.9 per million women per year is reported (5). PFTC is highly aggressive and associated with poor survival (1,6). Not much is known about the molecular and cytogenetic properties of this tumor type. Crude DNA content measurements have revealed that most PFTCs exhibit distinctly aneuploid cell populations (7,8). Overexpression of the tumor suppressor gene p53 has been reported in 50 to 60% of PFTCs (7,9,10); polymerase chain reaction (PCR)-single strand conformation polymorphism has confirmed p53 gene alterations in a high proportion of these immunopositive tumors (10). Only a few reports on chromosomal abnormalities in PFTCs exist because of its rarity and the technical difficulties in karyotype analysis with chromosome banding techniques. Chromosomally abnormal PFTC has been described in two case reports, but

From the Genome Technology Branch, National Human Genome Research Institute, National Institutes of Health, Bethesda, Maryland (K.H., E.S., T.R.), Department of Gynaecological Oncology, Radiumhemmet (A.-C.H.), and Department of Tumor Pathology, Karolinska Hospital and Institute, Stockholm, Sweden (H.B., C.S., G.A.), and the Department of Molecular Microbiology and Immunology, Johns Hopkins University School of Hygiene and Public Health, Baltimore, Maryland, (K.S.), U.S.A.

Address correspondence and reprint requests to Dr. Kerstin Heselmeyer, National Human Genome Research Institute, National Institutes of Health, 49 Convent Drive, Building 49, Room 4C36, Bethesda, MD 20892-4470.

detailed information about the aberrations were not reported because banding techniques were not applied in these studies (11,12). A third tumor described by Tharapel et al. (13) showed a normal karyotype, and Bardi et al. (14) reported a PFTC with complex chromosome aberrations similar to those in adenocarcinomas of other organs, including the ovaries and uterus.

Comparative genomic hybridization (CGH) is a molecular cytogenetic screening technique that makes it possible to identify chromosomal imbalances by analyzing genomic DNA (15). In a CGH experiment, differentially labeled tumor DNA and normal reference DNA are hybridized on normal metaphase chromosomes to map chromosomal copy number changes without tumor cell culture. Another important advantage is the possibility to analyze archival, formalin-fixed, and paraffin-embedded material (16,17). We used CGH on tumors microdissected from 50-μm sections prepared from routine histopathologic formalin-fixed and paraffin-embedded specimens to identify recurrent gains and losses in PFTC. In the same material, DNA ploidy measurements, screening for human papillomavirus (HPV) genomes, and immunohistochemical detection of the proliferation activity and the expression of the p53 tumor suppressor gene and the cyclin-dependent kinase inhibitor p21/WAF1 were performed.

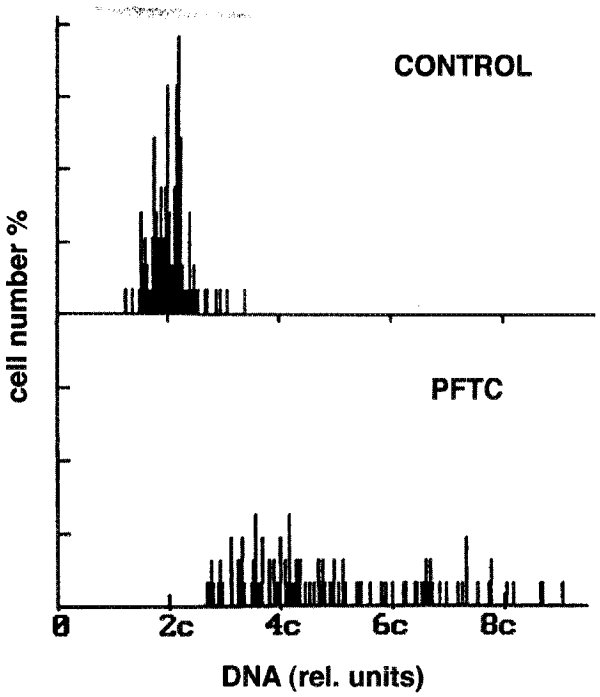

FIG. 1. Diploid DNA distribution pattern in normal human cells (control) and aneuploid DNA histogram typical for primary fallopian tube carcinomas.

MATERIALS AND METHODS

Tissue Samples

Twelve tumors were collected at the Department of Gynaecological Oncology, Radiumhemmet, Stockholm, between 1989 to 1996. The slides were reviewed at the Department of Pathology, Karolinska Hospital, Stockholm, and each case was staged according to FIGO (International Federation of Gynecology and Obstetrics) (18), based on the records and histopathologic results. On reexamination, the diagnostic criteria of Hu et al. (19) and Sedlis (20) were followed. Patient age, tumor staging, and the histologic features are presented in Table 1. Consecutive sections were cut from all formalin-fixed, paraffin-embedded samples. The tissue sections were used for DNA ploidy measurements (thickness 8 μm), immunohistochemistry (4 μm), and tumor DNA extraction (50 μm). The last section (4 μm) was stained with hematoxylin and eosin for confirmation of the presence of tumor. All tumor samples were obtained before treatment.

DNA Cytometry

DNA content measurements were performed by image cytometry on Feulgen-stained histologic sections as described (21). All DNA values were expressed in relation to the corresponding staining controls, which were given the value 2c, indicating the normal diploid DNA content. DNA histograms were interpreted according to Auer et al. (22). In the material investigated in this study, only aneuploid cases with > 20% of the cells exceeding 5c (AUER type IV histograms) were present. An example of a typical histogram is demonstrated in Figure 1, along with a histogram showing the DNA pattern of a normal diploid cell population.

Immunohistochemistry

Proliferative activity was determined with an antibody (MIB1; Immunotech S.A., Marseille, France) directed against the Ki-67 antigen. The antibody distinguishes cells in G0 from cells that are in G1-S-G2-M phase of the cell cycle. p21/WAF1 expression was analyzed with the WAF1 antibody EA10 (Oncogene Sciences, Uniondale, NY), and p53 expression was determined with the DO1 antibody (Santa Cruz Biotechnology, Santa Cruz, CA). Immunohistochemistry was performed as previously described (23). Proliferation was evaluated as low (< 20% immunoreactive tumor cells), moderate (20 to 40%), and high (> 40%). Frequencies of DO1 and WAF1 immunoreactive tumor cells were evaluated arbitrarily from "0" to "+++": 0, no immunopositive tumor cells; +, < 10%

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1.25, 1.5
14q13-q2

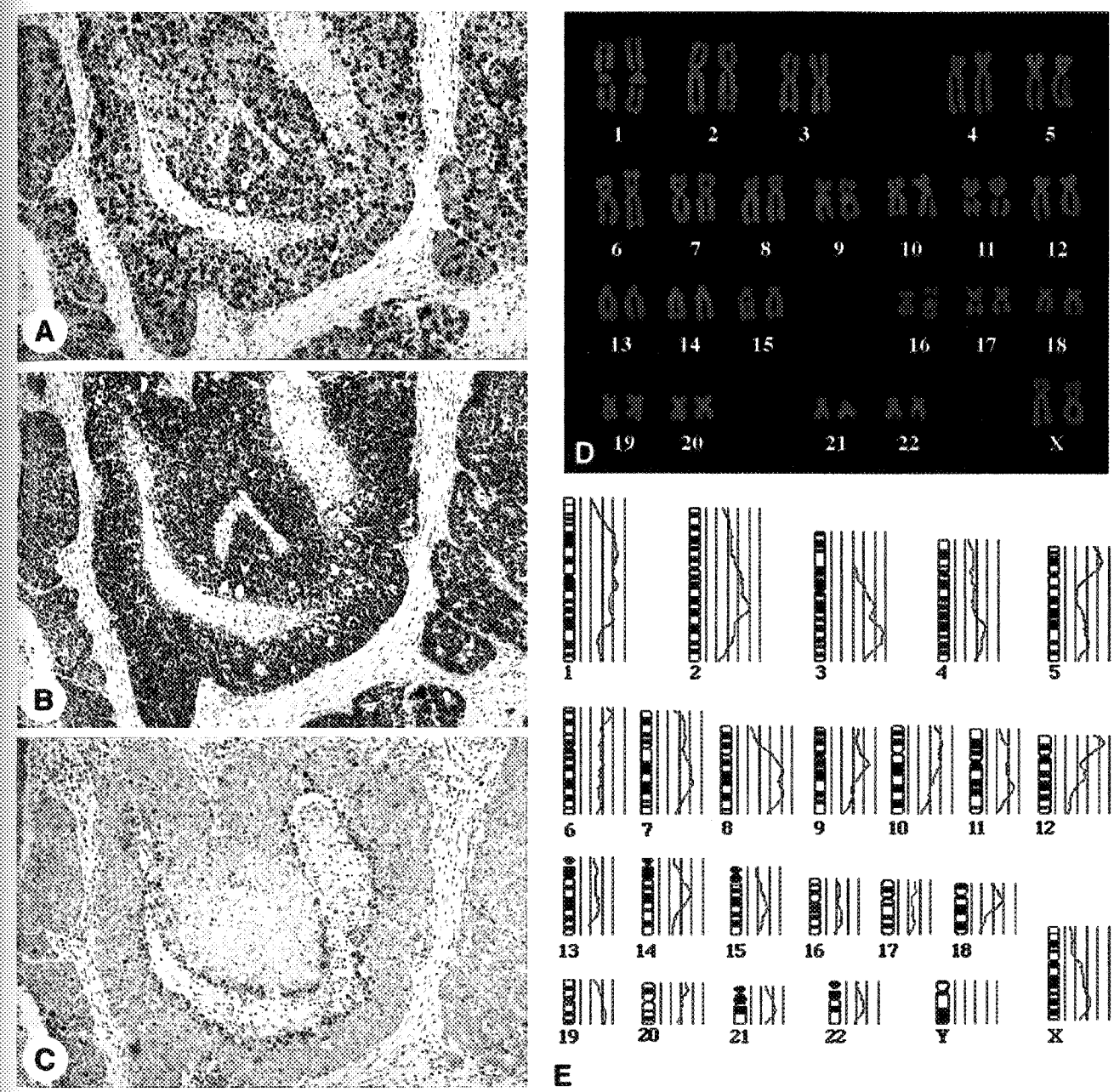

FIG. 2. Immunohistochemical detection of Ki-67 **(A)**, p53 **(B)**, and p21/WAF1 **(C)** in tissue sections of a primary fallopian tube carcinoma (case 8). Immunoreactive nuclei appear dark brown. The tissue was counterstained with hematoxylin. In this case, the proliferative activity was increased in 50 to 60% of the tumor cells **(A)**, approximately 90% of the tumor cells stained for p53 expression **(B)**, and few cells showed p21/WAF1 expression. **D:** A ratio image of case 8. Blue indicates a balance between tumor and test genomes, red reflects a loss of genetic material in the tumor DNA, and green shows regions that are gained in the tumor. **E:** The average ratio profile was computed on the basis of 10 ratio images. The vertical lines on the right side of the chromosome ideograms reflect different values of the fluorescence ratio between tumor and normal DNA. The black line demonstrates a balance between tumor and normal DNA (value 1.0). The red lines show the values 0.5 and 0.75 and the green lines show the values 1.25, 1.5, and 1.75. In this case, gains in copy number are present on 1p31-q31, 2q11.2-q24, 3q, 5p, 6pter-p22, 7q11.2-q31, 8q, 10pter-q21, 12p, 14q13-q21, and losses are present on 1pter-p36.1, 5q12-q23, 13q32-qter, 16, 18q21-qter and Xp.

TABLE 1. *Summary of the clinical data and results from MIB1, DO1, WAF1 staining, DNA ploidy measurements, HPV genotyping, and CGH in PFTCs*

Patient	List no.	Age, in yrs	Histology	Differentiation	Stage	MIB Ki67
1	FT 3	46	Serous papillary	Moderate	IA	M
2	FT 5	47	Serous papillary	Moderate	IIB	M
3	FT 6	54	Endometrioid	Moderate	IIB	L
4	FT 10	46	Serous papillary	Poor	IC	H
5	FT 11	51	Serous papillary	Poor	IIIB	H
6	FT 16	60	Serous papillary	Poor	IIIC	H
7	FT 18	66	Serous papillary	Poor	IIIC	H
8	FT 24	77	Serous papillary	Poor	IIIC	H
9	FT 25	56	Serous papillary	Poor	IV	M
10	FT 36	60	Serous papillary	Poor	IV	L
11	FT 45	80	Serous papillary	Poor	IIIB	H
12	FT 52	73	Serous papillary	Poor	IA	M

HPV, human papillomavirus; CGH, comparative genomic hybridization; PFTC, primary fallopian tube carcinoma.
The immunohistochemical results were evaluated semiquantitatively. MIB1 staining was divided into low (L), moderate (M), and high (H) proliferation activities. p53 and p21/WAF1 immunoreactivities were recorded as 0, +, ++, and +++. Only aneuploid (A) DNA histograms were retrieved. The CGH column shows the chromosomal aberrations detected in individual cases.

of tumor cells stained; ++, 10 to 50% tumor cells stained; and +++, strong staining in > 50% of the tumor cells (Table 1).

HPV Genotyping

HPV genomes in the purified DNA samples were screened by PCR with the MY09-MY11 L1 consensus primers used for amplification (24). We used one generic HPV probe and 25 type-specific probes to diagnose the HPV types in the PCR products (25). β-globin amplification was used as a positive control for evaluation of the adequacy of tumor DNA.

Comparative Genomic Hybridization

CGH was performed according to standard procedures (26). Tumor DNA was extracted from sodium-isothiocyanate-treated, formalin-fixed tissue sections (27). Nick translation was performed with bio-16-dUTP (tumor DNA) and digoxigenin-11-dUTP (reference DNA, prepared from a karyotypically normal female donor; 46, XX). Five hundred ng of each of the labeled genomes were combined and hybridized with an excess of Cot-1 DNA (25 μg) to metaphase chromosomes prepared from a karyotypically normal donor. Simultaneously, a mixture of PCR-generated alpha satellite probes labeled with Cy5-dUTP recognizing the centromeres of the chromosomes 4, 8, 14, 20, 22, and X was cohybridized to facilitate chromosome identification. The biotin-labeled tumor genome was visualized with avidin conjugated to FITC (Vector Laboratories), followed by biotinylated anti-avidin (Vector Laboratories)

TABLE 1. —*Continued.*

Patient	DO1 (p53)	WAF/p21	DNA ploidy	HPV	CGH
1	+	+	A	Negative	-2pter-p24,+2q31-q32,-2q33-qter,+3pter-p22,+3q26.2-qter,+5pter-p14,-6q23-qter, -7pter-p21,+8q21.1-qter,+pter-p22,+9q22-qter,-10q21-qter,-11pter-p15,-11q22-qter, -12q21.3,+13q31-qter,+14q31-qter,-16q22-qter,-17pter-q21,-21,-22
2	+++	0	A	Negative	-1pter-p34.3,+1p31.2-p22,+1q32,+2q22-q33,+3p13-qter,+4p14-q32,+5p14-q23, +6p23-p11.2(++6p22.3),-6q,+7q21-q31,-8p,+8q(++8q22-ter),+9p,-10q26, +11q14-q23.2,+12p,+12q15-q21,-12q24.3-qter,++13q21-qter,+14q12-q21,-14q32,-16, ++18q11.2-q22.1,-18q22.3-qter,-19q, -21q22.2-qter,-22,-Xq21-qter
3	0	+	A	Negative	+1q,+8,+12,+16,-21,+X
4	+++	+	A	Negative	+1p31,+1q,-2pter-p22,+2q23-q34,+3p13-qter(++3q26.1-qter),+4p,-4q32-qter,+5p, +6q12-q22,-6q23-qter,-8pter-p21,+9p,-10q23-qter,+11p,+11q14-q23.2,+12p, -13q12-q14,+14q21,+15q25-qter,-16q,-17pter-q21,-18p,+20p,-22,-Xq25-qter
5	+++	0	A	Negative	-1pter-p34.2,+1p31-p21,+1q25-qter,+2p21-p12,+2q24-q35,+3p13-qter,+5p,-5q31-qter, -6p21.3-p11.2,-6q25-qter,+7,-8pter-p21,+9p,-9q,+10p12-q21,+11p14-p12, +11q14.2-q23.2,+12pter-q15,-12q21-qter,-13q12-q14,+13q21-qter,+14q12-q22, -15q22-qter,-16,-17pter-q22,-19q,-22
6	+++	0	A	Negative	+1q42-qter,+2q22-q33,++3q,+4p15.1-q28,+5p,+6p22,+6q12-q24,-6q26-qter,+7q21-q22, +8q,+10p,-10q25-qter,+14q12-q21,-14q24-qter,-17p,-18q21-qter,+Xq
7	+++	0	A	Negative	+1q,+2,+3q22-qter,+5p,+6p,+6q25-qter,+7p15-p11.2,+7q21-q31,+8q13-q21.2, +13q31-qter,-14q31-qter,+15q21-qter,-16q,+17q,+18q11.2-q21,-22,-X
8	+++	+	A	Negative	-1pter-p36.1,+1p31-q31,+2q11.2-q24,++3q,+5p,-5q12-q23,+6pter-p22,+7q11.2-q31,+8q, +10pter-q21,++12p,-13q32-qter,+14q13-q21,-16,-18q21-qter,-Xp
9	+++	+	A	Negative	++1q42-qter,+2p13-q32,-3p,+3q(++3q26.2-qter),-4q32-qter,-5q11.2-q21,+6pter-p22, -7q31,+7q32-qter,-8p,+8q(++8q22-qter),+10p13-p11.2,-11pter-p14,+11q,-13q21-qter, +16p,+18p11.2-q21,-18q22-qter,+19q,+20,-X
10	+++	0	A	Negative	-1pter-p35,+1q,+2p23-p14,+3q,+6p23-p12,+7q,+8p,-9p,-11q23-qter,++12p,+12q12-q22, +13q12-q31,+18pter-q12,-18q21-qter,+19q13.1,+20q,+21,-X
11	+++	+	A	Negative	+1p22-q31,+2q22-q32,++3p14-qter,-4p,+5p14-q12,-5q31-qter,+6p,-6q26-qter, +7q11.2-33(++7q21),-8p,+8q,+9,+10p13-p11.2,-11q24-qter,++12p,+12q12-q22, -13q31-qter,+14q21,-14q31-qter,-15,-16,-17p,-18q21-qter,+20p,-21q22,-22q13
12	+++	0	A	Negative	-1pter-p36.1,+1q31.2-qter,+2q23-qter,+3p12-qter(++3q26.2-qter),+4q12-q21,+6q12-q25, +7q11.2-q32,-8p,+8q23-qter,-12q21-qter,+15q26,-16q,+20p,-22,-X

and another layer of FITC-conjugated avidin. The digoxigenin-labeled reference DNA was detected with a mouse anti-digoxin antibody (Sigma, St. Louis, MO), followed by a rabbit anti-mouse antibody conjugated to TRITC (Sigma) and a goat anti-rabbit antibody conjugated to TRITC (Sigma). Chromosomes were counterstained with DAPI to generate a G-like banding pattern.

Gray scale images of the DAPI counterstain, the FITC-labeled tumor DNA, the TRITC-labeled reference DNA, and the Cy5 labeled centromeres from 12 to 15 metaphases from each hybridization were acquired with a cooled CCD camera (CH250; Photometrics, Tucson, AZ) connected to a Leica DMRXA microscope equipped with fluorochrome specific optical filters TR1, TR2, TR3, and TR5 (Chroma Technology, Brattleboro, VT). Quantitative evaluation of the hybridization was achieved with the Applied Imaging Cytovision system (Applied Imaging International, Tyne & Wear, United Kingdom). Average ratio profiles were computed as the mean value of at least 8 metaphase ratio images (Fig. 2D), and used to identify chromosomal copy number changes in all cases.

RESULTS

The clinical data and the results of the different analyses are summarized in Table 1.

CGH Analysis

Figure 2 shows the visual summary of the immunohistochemical and the CGH data for case 8. The CGH result is presented as a ratio image of a representative metaphase cell (Fig. 2D) and as an average profile of 10 metaphases of the same case (Fig. 2E). The ratio image

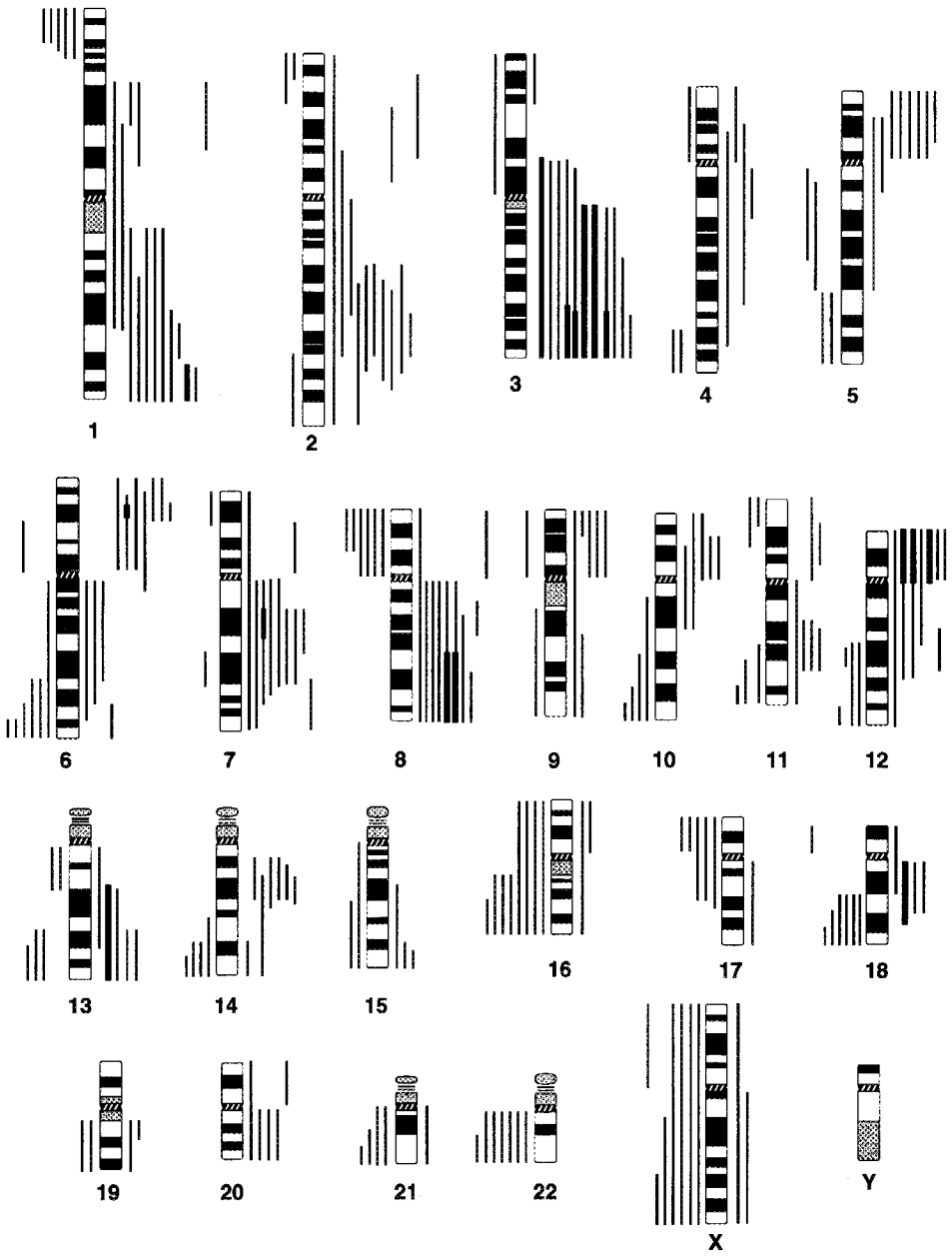

FIG. 3. Karyogram of gains and losses in 12 cases of primary fallopian tube carcinomas. Lines on the right side of the ideograms reflect copy number increases, lines on the left represent copy number decreases, and bold lines symbolize high level copy number increases (amplifications).

reflects gains and losses by pseudocolors. Blue indicates a balance between tumor and test genomes, red reflects a loss of genetic material in the tumor DNA, and green shows regions that are gained in the tumor. The final step in a quantitative fluorescence measurement requires the calculation of average ratio profiles along the chromosomal axis based on data from at least 8 metaphase spreads. The average ratio profiles were used for mapping of copy number changes in all instances.

Chromosomal aberrations were detected in all 12 tumors; the average number of chromosomal aberrations per case was 19.7. Despite the high number of chromosomal aberrations, a recurrent and specific pattern emerged in the fallopian tube carcinomas. The most frequent copy number change was a gain of chromosome arm 3q, which occurred in 11 of 12 cases (92%). In six cases, the gain of 3q was present as a high level copy number increase (amplification). The consensus region of 3q amplification was mapped to 3q26.2-qter. Genetic material was also frequently gained on chromosome 1q (in 11 cases), 2q (in 10), 7q (in 9), 8q (in 9), 5p (in 8), 6p (in 7), 12p (in 7), and 14q (in 6). Three of the 12 cases showed a whole-arm amplification on 12p. Regional high level copy number increases (amplifications) were

mapped in one case to 6p22.3, 8q22-qter, 13q21-qter, and 18q11.2-q22.1, and in another case to 1q42-qter, 3q26.2-qter, and 7q21. Frequent losses occurred on chromosome arms 16q (in 8 cases), 22q (in 7), 6q (in 6), 8p (in 6), 18q (in 6), Xq (in 6), 1p (in 5), and 17p (in 5).

Of note, there was a difference between the only carcinoma with endometrioid histology and the 11 serous papillary carcinomas. Although all of the serous papillary carcinomas revealed high numbers of chromosomal aberrations (average of 20.9 aberrations per case), the endometrioid carcinoma showed only 6 aberrations that were either whole chromosome or whole chromosome arm aberrations. A summary of the chromosomal gains and losses found in the 12 PFTCs is presented in Figure 3.

DNA Ploidy Measurement

DNA ploidy was measured on consecutive tissue sections by image cytometry. Image cytometry complements the CGH analyses by providing information of the DNA ploidy that cannot be gained from CGH experiments alone. None of the tumors showed a normal diploid DNA content. All cases showed distribution patterns with highly aneuploid cell populations; > 20% of the cells exceeded the 5c value. The results are presented in Table 1.

HPV Genotyping

All samples could be analyzed successfully with a primer for the β-globin gene as a control. All 12 samples tested negative for HPV genomes.

Immunohistochemistry

Six samples showed high proliferative activity, four showed moderate activity, and two showed low activity. The only case (case 3) that did not show p53 immunoreactivity was the endometrioid carcinoma; the 11 serous papillary carcinomas exhibited p53 immunoreactivity. In 10 of the 12 cases, > 50% of the tumor cells stained positively. Immunoreactivity for p21/WAF1 was weak or negative for all cases. The focally immunoreactive (staining of < 10% of the tumor cells) and the p53 nonimmunoreactive case showed weak p21/WAF1 immunoreactivity. Examples of the immunohistochemical findings (case 8) are provided in Figures 2A to C. The data are summarized in Table 1.

DISCUSSION

This study summarizes the results of a combined phenotype–genotype analysis of 12 PFTCs. Using CGH as a molecular cytogenetic screening test for copy number changes, we identified a recurrent pattern of chromosomal aberrations in these tumors. DNA content measurements revealed aneuploid distribution patterns for all 12 carcinomas. Proliferation activities were elevated for most cases and highly elevated for half of the carcinomas. All cases except one exhibited p53 immunoreactivity. In 10 of the 12 cases, > 50% of the tumor cells stained positively. Immunoreactivity for p21/WAF1 was weak or negative for all cases. No HPV genomes were detected in any of the samples.

Our CGH data indicate that PFTCs are genetically complex. The average number of copy alterations (ANCA) was 19.7 per tumor, which is very high compared to other cancer types; small cell lung cancer has an ANCA of 13.0 (28), glioblastomas have an ANCA of 8.3 (29), advanced cervical carcinomas have an ANCA of 8.2 (30), aneuploid breast cancers have an ANCA of 6.8 (27), colorectal adenocarcinomas have an ANCA of 5.6 (23), and anal squamous cell carcinomas have an ANCA of 4.8 (31). Every chromosome arm was involved in DNA sequence copy number changes. Despite the high number of copy alterations, changes were clustered at certain regions, revealing a recurrent nonrandom pattern of chromosomal aberrations in PFTCs. A gain of chromosome arm 3q (11 of 12 tumors) was the most common aberration observed. In six cases, the gain of 3q was present as high level copy number increased. The smallest amplicon could be mapped to the region 3q26.2-qter.

Frequent copy number gains and localized amplifications on the long arm of chromosome 3 have been observed in a number of solid tumor types, including ovarian cancer, small cell lung cancer, cervical cancer, and squamous cell carcinoma of the head and neck, lung, and anus (26,28,30–38). In contrast, adenocarcinomas of the breast, colon, and lung rarely show these changes (23, 27,38). These observations indicate that the 3q gain is a very frequent and consistent finding in squamous cell carcinoma. However, the frequent 3q gain in ovarian and tubal carcinomas shows that 3q also plays an important role in some adenocarcinomas. The human telomerase RNA gene (hTR)—a promising candidate gene—has recently been mapped to chromosome 3q26.3 (39). Soder et al. (39) found increased copy numbers of the hTR locus in 97% of the cervical, head and neck, and lung carcinomas they investigated and an amplification of the hTR locus could be demonstrated in 4 of the tumors.

Other characteristic copy number increases were mapped to chromosomes 1q, 2q, 7q, 8q, 5p, 6p, 12p, 14q, and 20q in our study. Consistent losses, indicative of chromosomes where tumor suppressor genes reside, were mapped to chromosomes 16q, 22q, 6q, 8p, 18q, Xq, 1p and 17p. Of note, Bardi et al. (14), in the only available banding analysis of a PFTC with chromosome abnormalities, reported several gains and losses which are

in agreement with the pattern we observed, specifically a loss of 1pter-p34, an isochromosome 8q and a loss of chromosomes 16, 17, 18, and 22. However, a 3q-gain—the most frequent aberration in our material—was not observed for that case (14).

The pattern of chromosomal aberrations in PFTCs is in many respects similar to those reported for other gynecologic cancers. Gain of 1q, 3q, 5p, 6p, 8q, and 20q and loss of 18q and X also has been frequently observed in ovarian (34,37,40) and cervical cancer (30). Additionally, ovarian cancers have also shown frequent gain of 12p and frequent loss of 16q and 17p. For some of these common regions of aberrations, candidate oncogenes and tumor suppressor genes are known. The amplification of 8q material could be related to C-myc. Baker et al. (41) found that C-myc amplification is a common finding in advanced-stage ovarian carcinomas. We observed an 8q gain and a concomitant 8p loss in 3 of the 12 PFTCs, indicating an isochromosome 8q formation. Isochromosome formations of 8q have also been observed by classical banding analysis in ovarian tumors (42) and primary cervical carcinomas (43).

A gene which might be potentially involved in the frequent gain of 12p and the 12p amplification observed in three PFTCs is the K-ras gene that maps to 12pter-p12. Mizuuchi et al. (44) detected K-ras mutations in seven of eight PFTCs. It has also been shown that K-ras may be amplified in approximately 5 to 10% of epithelial ovarian cancers (45).

We observed a loss of 17p in 5 of the 12 PFTCs. The p53 tumor suppressor gene resides on 17p12-p13.3. Our additional p53 immunohistochemical studies showed focal positivity in one of these cases and strong positivity in the other four. The positive immunohistochemical assay and the observed loss of 17p could indicate a loss of one allele and a concomitant mutation of the other allele.

The smallest consensus region we observed for the 18q loss was 18q22.3-qter which is beyond the DCC gene locus 18q21. Arnold et al. (37) observed 18q loss in advanced ovarian cancer with a consensus region on 18q22-qter. These findings are in accordance with the loss of heterozygosity studies of ovarian adenocarcinomas (46) in which a consensus region between the genetic loci D18S5 and D18S11 was found, indicating that another, more distally localized tumor suppressor gene may be inactivated in ovarian and tubal carcinomas.

The immunophenotype we obtained for the PFTCs is similar to the pattern we observed for colorectal carcinomas (23). Ten of the 12 PFTCs showed p53 immunoreactivity in > 50% of the tumor cells although their p21/WAF1 expression was low or negative, a finding suggesting mutated p53 protein. Runnebaum et al. (47) and Zheng et al. (10) observed p53 mutations in ~ 60% of their PFTCs, which is similar to our immunohistochemical data.

Of note, the only p53-negative case was the only case that belonged to a different histologic subgroup. Although 11 of the 12 PFTCs were classified as serous papillary tumors, this exceptional case belonged to the endometrioid subgroup. Navani et al. (48) suggested that endometrioid tubal carcinomas are less aggressive and have a generally favorable prognosis; however, Hellström et al. (49) did not report a better survival for patients with endometrioid PFTC. The CGH results for our endometrioid carcinoma differed noticeably from the 11 serous PFTCs; only 6 aberrations were observed as compared to an average of 20.9 in the serous tumors. All of the six aberrations observed in the endometrioid carcinoma were either whole chromosome or whole chromosome arm aberrations. Remarkably, this is also the only case which did not show a gain of the long arm of chromosome 3. A larger study is necessary to determine whether a difference between the genotypes of serous papillary and endometrioid PFTC exists.

We previously demonstrated frequent 3q gains in HPV-associated tumor types, such as cervical and anal carcinoma (26,30,31), that suggested a possible correlation between 3q gain and HPV infection. In this study, we observed that PFTCs that tested negative for HPV sequences showed a high frequency of 3q gains, indicating that the 3q-gain occurs independently from the presence of HPV DNA in PFTC.

The data of this study reveal that PFTCs are characterized by an extremely unstable genotype demonstrated by highly scattered DNA ploidy patterns and a remarkably high number of copy alterations determined by CGH. A specific pattern of chromosomal aberrations showed similarities to the CGH patterns obtained for other gynecologic cancers. 3q-gain was the most frequent aberration and could be observed in all of the serous papillary PFTCs. No HPV infection was detected and most of the cases showed strong p53 immunoreactivity, suggesting p53 mutation. These findings are supported by the observation that PFTC is a highly malignant cancer with a poor prognosis. The ANCA observed in PFTC is exceptionally high and suggests a strong correlation between the CGH ANCA and the malignant behavior of the tumor (27,50).

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